Pergamon

Tetrahedron 55 (1999) 1793–1798

TETRAHEDRON

A New Efficient Procedure for the Reduction of Δ^7 Ergostane Derivatives

Carme Brosa* **and Carlos Rodríguez-Santamarta**

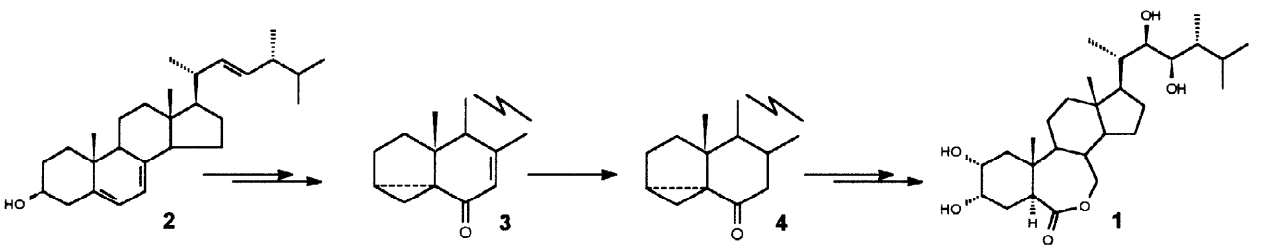
Department of Organic Chemistry, Institut Químic de Sarrià CETS; Universitat Ramon Llull, Via Augusta 390, 08017- Barcelona, Spain.

Received 21 September 1998; revised 17 November 1998; accepted 10 December 1998

Abstract: The reduction of Δ^7-6-keto-ergostane derivatives with $Na_2S_2O_4$ in the presence of a phase transfer catalyst is performed and the effect of experimental conditions is discussed. This represents a safer and economic way for the synthesis of brassinosteroid analogs.

In the field of brassinosteroids, potent natural plant growth promoters,[1] most synthetic efforts are focused on generating procedures to obtain the active compounds in an easier and economic way. Among the active natural brassinosteroids, 24-epibrassinolide (**1**) has been widely evaluated on field trials in several crops. The results obtained have successfully demonstrated the suitability of this steroid for improving crop production.[2]

Although several synthetic strategies of 24-epibrassinolide (**1**) have been developed, most of them involve a reduction of the Δ^7 double bond present in the ergosterol (**2**) starting material.[3-10] Thus, one of the general procedures to prepare 24-epibrassinolide (**1**) is represented in Scheme 1 where the ketone **4** is obtained from the enone **3** in a yield of 80%.[3,8]



Scheme 1

* e-mail. brosa@iqs.url.es fax. 34 93 2056266

0040-4020/99/$ - see front matter
PII: S0040-4020(98)01191-0

This reduction, as in other sterically hindered enones, has usually been accomplished by treatment with lithium in liquid ammonia.[11] However, this procedure presents serious drawbacks mainly for large-scale preparation:

i) the difficulty of reaction control to avoid useless overreduction products;
ii) the uneconomic conditions derived from the low temperature (-78°C) required;
iii) the security measures needed for working with lithium in liquid ammonia pre-distilled over sodium, and
iv) the exhaustive control of residues to minimise the environmental impact.

Sodium dithionite, used as a bleaching agent,[12] appears as a commercial reducing agent to accomplish selective enone reduction. Therefore, it could be considered as a suitable candidate to overcome the difficulties cited above.

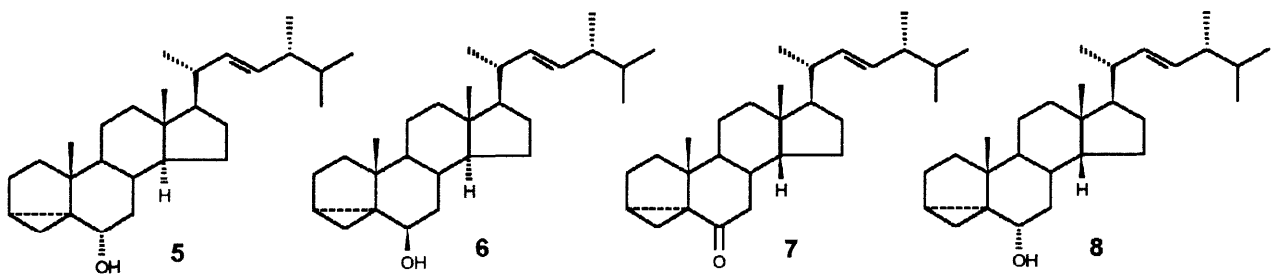
In this paper we present the opportunity to use this reagent for the reduction of Δ^7-ketosteroids in a satisfactory way.

Results and discussion:

Following to the literature,[13] initial experiments were performed in monophasic systems, obtaining unsatisfactory results. Thus, when the reaction was carried out in an aqueous medium with the presence of dioxane as cosolvent the starting material remained unreacted. On the other hand, using dimethylformamide to carry out the reaction at a higher temperature a complex crude was obtained.

The use of a phase transfer catalyst (PTC) such as Aliquat® 336 following the procedure of Camps *et al.*[14] and Akamanchi *et al.*[15] for the reduction of ketones, enones and conjugated dienoic carboxylic acids and esters furnishes the desired ketone **4** as main product.

Otherwise, minimal variations of experimental conditions affect the reaction behaviour obtaining either new characterised overreduction products (**5** and **6**), isomerization ones (**7**) or those resulting from both processes (**8**).



Scheme 2

Table 1 shows the results obtained in a toluene/water system by modifying different factors: the amount of PTC, the basic conditions, the $[Na_2S_2O_4]$/[**3**] ratio and the path of reagent addition.

Hence, whereas a deficiency of base gives a substantial amount of unreacted enone **3** (Entry 1), an excess of it leads to the formation of isomerization (**7**) and isomerization-overreduction products (**8**) (Entry 3).

Trace amounts of PTC as well as reducing agent ratio of 1.5 involving long time reaction increases the amount of **8** (Entry 4 and 6).

Taking into account the best experimental conditions encountered -the molar relation of $NaHCO_3$ (**8**), Aliquat® (9) and $Na_2S_2O_4$ (9)- an increase in the yield of the target molecule **4** is obtained with a slow reagent addition (Entry 2). Thus, with this procedure a crude product with a content of 80% in **4** is obtained (72% after chromatographic purification). However, 10% of undesired product **8** is obtained and 10% of the starting material **3** remains unchanged.

Table 1. Reaction conditions for the reduction of enone **3**.

Variable	Entry	$NaHCO_3$[#]	Aliquat® 336[#]	$[Na_2S_2O_4]$[#]	Time /h	**4**[‡]	**5**+**6**[‡]	**7**[‡]	**8**[‡]	**3**[‡]
	1	1	9	1.5 x 6	6	25				75
$NaHCO_3$	2	8	9	1.5 x 6	2	80			10	10
	3	18	9	1.5 x 6	2	65		10	20	5
	4	8	traces	1.5 x 6	7	25			50	25
Aliquat®336	2	8	9	1.5 x 6	2	80			10	10
	5	8	18	1.5 x 6	2		90			10
$[Na_2S_2O_4]$	6	8	9	1.5	8	40			40	20
	2	8	9	1.5 x 6	2	80			10	10
Reagent	7	8	9	9	2	70				30
addition	2	8	9	1.5 x 6	2	80			10	10

[#] mmol / mmol of **3**. [‡] Yields calculated by HPLC.

An excess of PTC gives the formation almost quantitatively of the overreduced alcohols **5** and **6** (Entry 5), which are useful to be easily transformed into the desired ketone **4** after a re-oxidation step.

Therefore, the overreduction of enone **3**, followed by a Jones re-oxidation, without intermediate purification, afforded, after chromatographic purification, the ketone **4** in a 82 % yield, recovering a 10% of unreacted enone **3**.

This two-step procedure presents high yields, implying easy reaction control. Therefore, this represents a convenient alternative way for the synthesis of ketone **4**.

Conclusions.

The use of $Na_2S_2O_4$ in the presence of a phase transfer catalyst represents a more convenient, safer and economic way for the reduction of Δ^7-6-keto ergostane derivatives, a key step for the synthesis of brassinosteroid analogs.

Experimental section:

Melting points were determined on a Gallenkamp and on a Büchi 530 instruments and are uncorrected. IR spectra were obtained on a Perkin -Elmer 683 spectrometer. ^{1}H-NMR and ^{13}C-NMR spectra were recorded on a Varian-Gemini 300 (300MHz) spectrometer in $CDCl_3$ solutions. Chemical shifts (δ) are given in ppm with Me_4Si as internal standard. The multiplicity of the signals in the ^{13}C-NMR was determined using the sequence Distorsionless Enhancement Polarization Transfer (D.E.P.T.). Mass spectra (electronic impact (EIMS), m/z) were run on a Hewlett-Packard 5985-A spectrometer and mass spectra (chemical ionization, (CIMS) m/z) were run on a VG autoSPEC spectrometer using methane as the carrying gas. HPLC chromatography was run on a Waters 600E chromatograph, an ultraviolet Waters detector (λ=210 nm), a Waters-Millipore Nova-Pak RP-18 (ϕ_i=3.9 mm, μ=4μm, L=15 mm) column and CH_3CN/H_2O 60:40 as mobil phase. Flash chromatography was performed on 230-400 mesh MN silica gel.

General reduction procedure: A solution of (*22E*)-3α,5-cyclo-5α-ergosta-7,22-dien-6-one (**3**) (1 mmol, 395 mg) in toluene (40 ml) was added to water (40 ml) containing $NaHCO_3$ (8 mmol, 677 mg) and Aliquat® 336 (9 mmol, 4 ml) under argon. Sodium dithionite (1.5 mmol, 250 mg) was added and reaction mixture was stirred at reflux temperature for 2 hours, during which time additional sodium dithionite was added in five portions of 250 mg each one. The reaction mixture was cooled to room temperature and cold water was added until the solution became clear. After the usual work-up (Cyclohexane, Cy) and chromatographic purification (Cy/AcOEt 80:1) 290 mg of (*22E*)-3α,5-cyclo-5α-ergost-22-en-6-one (**4**) was isolated (72% yield) as a white solid; **mp**: 167-169°C (lit[16] 168-169°C) **IR** ν^{CHCl_3} cm^{-1}: 1690, 1455, 1365, 1290, 960. **^{1}H-NMR** (300 MHz, $CDCl_3$): δ 5.26-5.11 (2H, m, H-C22 + H-C23), 2.49-2.37 (1H, m, βH-C7), 2.06-2.02 (1H, m, αH-C7), 1.02 (3H, d; J=6.9Hz, 21-CH_3), 1.01 (3H, s, 19-CH_3), 0.91 (3H, d, J=6.9Hz, 28-CH_3), [0.84 (3H, d, J=6.9Hz) + 0.82 (3H, d, J=6.9Hz)] [26-CH_3 + 27-CH_3], 0.73 (3H,s,18-CH_3), 0.72 (1H, dd, J=5.1, 5.1Hz, αH-C5); **^{13}C-NMR** (75 MHz, $CDCl_3$): 209 C6, 135.5 C22, 132.0 C23, 57.1 C14, 56.0 C17, 46.8 C10, 46.3 C5, 46.1 C9, 44.8 C7, 42.8 C24, 42.6 C13, 40.1 C20, 39.6 C12, 35.3 C8, 34.8 C3, 33.5 C1, 33.1 C25, 28.4 C16, 25.9 C2, 24.0 C15, 22.8 C11, 20.9 C21, (19.9 + 19.6) (C26 + C27), 19.7 C19, 17.6 C28, 12.2 C12, 11.6 C4. **MS** (EI): 396 (M^+), 381, 353, 299, 298, 283, 271, 269, 149, 69, 55.

Two-step procedure: A solution of (*22E*)-3α,5-cyclo-5α-ergosta-7,22-dien-6-one (**3**) (1 mmol, 395 mg) in toluene (40 mL) was added to water (40 mL) containing $NaHCO_3$ (8 mmol, 677 mg) and Aliquat® 336 (10 mmol, 8 mL) under argon. Sodium dithionite (1.5 mmol, 250 mg) was added and reaction mixture was stirred at reflux temperature for 2 hours, during which time additional sodium dithionite was added in five portions of 250 mg each one. The reaction mixture was cooled to room temperature and cold water was added until the solution became clear. After the usual work-up (Cy), Aliquat®336 was removed after filtration over silica. 390 mg of a crude containing (*22E*)-3α,5-cyclo-5α-ergost-22-en-6α-ol (**5**) and (*22E*)-3α,5-cyclo-5α-ergost-22-en-6β-ol (**6**) (90 % yield by HPLC) was obtained. Jone's reagent was added dropwise to a solution of the above crude in 8 mL of acetone (distilled from $KMnO_4$), at 0°C, until the mixture remained reddish.The reagent excess was removed by adding *i*-PrOH and the solution was filtered over celite. After removing the solvent under reduced pressure and usual work-up ($CHCl_3$) a crude of 410 mg was obtained. The chromatographic purification (Cy/AcOEt 80:1) afforded 325 mg of 3α,5-cyclo-5α-ergost-22-en-6-one (**4**) (82% yield from **3**) and 40 mg of unreacted enone **3**.

(*22E*)-3α,5-Cyclo-5α-ergost-22-en-6α-ol (**5**): white solid

mp: 83-84°C. **IR** ν^{CHCl_3} cm^{-1}: 3600-3100, 1450, 1370, 1000-1050, 960. **^{1}H-NMR** (300 MHz, $CDCl_3$): δ 5.26-5.11 (2H, m, H-C22 + H-C23), 3.90 (1H, dd, J=11.4, 4.2Hz, βH-C6), 1.00 (3H, d, J=6.3Hz, 21-CH_3), 0.91 (3H, s, 19-CH_3), 0.91 (3H, d, J=6.3Hz, 28-CH_3), 0.83 (3H, d, J=6.9Hz, 27-CH_3), 0.82 (3H, d, J=6.6Hz, 26-CH_3), 0.70 (3H, s, 18-CH_3), 0.61 (1H, dd, J=8.1, 4.5Hz, H_1-C4), 0.25 (1H, dd, J=4.2, 4.2Hz, H_2-C4); **^{13}C-NMR** (75 MHz, $CDCl_3$): 135.8 C22, 131.7 C23, 67.2 C6, 56.3 C14, 56.1 C17, 47.7 C9, 44.9 C10, 42.8 C24, 42.6 C13, 40.2 C7, 40.2 C20, 40.0 C12, 39.8 C5, 34.9 C8, 33.1 C25, 32.8 C1, 28.5 C16, 25.0 C2, 24.2 C15, 23.1 C11, 20.9 C21, 19.9 C27, 19.6 C26, 18.7 C3, 17.9 C19, 17.6 C28, 12.3 C18, 6.6 C4.**CIMS (h.r.)** m/z:$[M+1]^+$ 398.355 (calc.398.355).

(*22E*)-3α,5-Cyclo-5α-ergost-22-en-6β-ol (**6**): white solid

mp: 81-82°C. **IR** ν^{CHCl_3} cm^{-1}: 3600-3100, 1450, 1370, 1000-1050, 960. **^{1}H-NMR** (300 MHz, $CDCl_3$): δ 5.26-5.11 (2H, m, H-C22 + H-C23), 3.26 (1H, dd, J=2.1, 2.1Hz, αH-C6), 1.06 (3H, s, 19-CH_3), 1.00 (3H, d, J=6.3Hz, 21-CH_3), 0.91 (3H, d, J=6.3Hz, 28-CH_3), 0.83 (3H, d, J=6.9Hz, 27-CH_3), 0.82 (3H, d, J=6.6Hz, 26-CH_3), 0.74 (3H, s, 18-CH_3), 0.52 (1H, dd, J=3.9, 3.9Hz, H_1-C4), 0.29 (1H, dd, J=8.4, 5.1Hz, H_2-C4); **^{13}C-NMR** (75 MHz, $CDCl_3$): 135.8 C22, 131.7 C23, 73.8 C6, 56.6 C14, 56.2 C17, 47.7 C9, 42.8 C13 + C10, 42.6 C24, 40.2 C20, 40.1 C7, 38.9 C5, 37.1 C12, 32.8 C1, 33.1 C25, 29.8 C8, 28.6 C16, 25.0 C2, 24.2 C3, 24.1 C15, 22.6 C11, 20.9 C21, 20.2 C19, 19.9 C27, 19.6 C26, 17.6 C28, 12.4 C18, 11.6 C4. **CIMS (h.r.)** m/z:$[M+1]^+$ 398.355 (calc.398.355).

(*22E*)-3α,5-Cyclo-5α,14β-ergost-22-en-6-one (**7**): white solid

mp: 144-147°C. **IR** ν^{CHCl_3} cm^{-1}: 1690, 1450, 1370, 1285, 1150. **^{1}H-NMR** (300 MHz, $CDCl_3$): δ 5.26-5.11 (2H, m, H-C22 + H-C23), 3.26 (1H, dd, J=2.1, 2.1Hz, αH-C6), 1.06 (3H, s, 19-CH_3), 1.00 (3H, d, J=6.3Hz, 21-CH_3), 0.91 (3H, d, J=6.3Hz, 28-CH_3), 0.83 (3H, d, J=6.9Hz, 27-CH_3), 0.82 (3H, d, J=6.6Hz, 26-CH_3), 0.74 (3H, s, 18-CH_3), 0.52 (1H, dd, J=3.9, 3.9Hz, H_1-C4), 0.29 (1H, dd, J=8.4, 5.1Hz, H_2-C4); **^{13}C-NMR** (75 MHz, $CDCl_3$): 135.8 C22, 131.7 C23, 73.8 C6, 56.6 C14, 56.2 C17, 47.7 C9, 42.8 C13 + C10, 42.6 C24, 40.2 C20, 40.1 C7, 38.9 C5, 37.1 C12, 32.8 C1, 33.1 C25, 29.8 C8, 28.6 C16, 25.0 C2, 24.2 C3, 24.1 C15, 22.6 C11, 20.9 C21, 20.2 C19, 19.9 C27, 19.6 C26, 17.6 C28, 12.4 C18, 11.6 C4. **CIMS (h.r.)** m/z:$[M+1]^+$ 396.338 (calc.396.339).

(*22E*)-3α,5-Cyclo-5α,14β-ergost-22-en-6α-ol (**8**): white solid

mp: 78-80°C **IR** ν^{CHCl_3} cm^{-1}: 3600-3100, 1450, 1370, 1230, 1045. **^{1}H-NMR** (300 MHz, $CDCl_3$): δ 5.26-5.11 (2H, m, H-C22 + H-C23), 3.90 (1H, dd, J=11.7, 3.9Hz, βH-C6), 1.04 (3H, s, 19-CH_3), 0.93 (3H, d, J=6.3Hz, 21-CH_3), 0.92 (3H, d, J=6.3Hz, 28-CH_3), 0.88 (3H, s, 18-CH_3), 0.84 (3H, d, J=6.9Hz, 27-CH_3), 0.83 (3H, d, J=6.6Hz, 26-CH_3), 0.60 (1H, dd, J=8.1, 4.5Hz, H_1-C4), 0.24 (1H, dd, J=4.2, 4.2Hz, H_2-C4); **^{13}C-NMR** (75 MHz, $CDCl_3$): 133.5 C22, 132.9 C23, 67.5 C6, 56.0 C14, 49.1 C9, 44.8 C10, 43.4 C13, 43.3 C24, 41.0 C7, 39.9 C5, (39.3 + 37.6) (C17 + C20), 36.7 C12, 33.2 C25, 32.9 C1, 32.8 C8, 25.2 C2, (24.8 + 23.3) (C15+C16), 23.1 C11, 22.0 C21, 20.1 C27, 19.7 C26, 19.1 C18, 18.8 C3, 17.7 C19, 17.6 C28, 6.7 C4. **CIMS (h.r.)** m/z:$[M+1]^+$ 398.353 (calc. 398.355).

Acknowledgements:

This work was supported by the Comissió Interdepartamental de Recerca i Innovació Tecnológica (CIRIT. QFN95-4716). We wish to thank Dr. F. Lafont from the Universidad de Córdoba for the HRMS spectra.

References:

1. Cutler, H.G.; Yokota, T. and Adam, G. *Brassinosteroids; Chemistry, Bioactivity & Applications (ACS Symp. Ser. 474)*, American Chemical Society, Washington, DC, **1991**.
2. Brosa, C.; Biological effects of brassinosteroids. *Biochemistry and function of sterols*, Ed. E.J. Parish, D. Nes. CRC Press, Inc., Boca Raton **1997**, Chap. 15, 201-218.
3. Thompson, M.J.; Mandava, N.; Flippen-Anderson, J.L.; Worley, J.F.; Dutky, S.R.; Robbinson, W.E.; Lusby, W. *J. Org. Chem.*, **1979**, *44*, 5002.
4. Anastasia, M.; Ciuffreda, P.; Fiechi, A. *J. Chem. Soc. Perkin Trans. I*, **1983**, 379.
5. Takatsuto, S.; Ikekawa, N. *Chem. Pharm. Bull (Tokyo)*, **1988**, *32*, 2001.
6. Zhou, W.; Zhou, Y.; Liang, B. *Synthesis*, **1989**, *6*, 426.
7. Ferrer, J.C.; Lalueza, R.; Saavedra, O.M.; Brosa, C. *Tetrahedron Lett.*, **1990**, *31*, 3941.
8. McMorris, T.C.; Patil, P.A. *J. Org. Chem.*, **1993**, *58*, 2338.
9. Khripach, V.A.; Zhabinski, V.N.; Ol'khovik, V.K.; Ivanova, G.I.; Zhermosek, E.V. *Zh. Org. Khim.*, **1994**, *30*, 1650.
10. Brosa, C.; Puig, R.; Comas, X.; Fernandez, C. *Steroids*, **1996**, *61*, 540.
11. Dryden, H.L.Jr.; Reduction of steroids by metal-ammonia solutions; *Organic Reactions in Steroids Chemistry*; Ed. Fried, J.and Edwards, J. A., Van Nostrand Reinhold Company, New York **1972**, *Vol.I*, 1.
12. See for instance: Baumgarte, U. *Textilveredlung* **1967**, *2*, 896.
13. Dhillon, R.; Singh, R.; Kaur, D. *Tetrahedron Lett.*, **1995**, *36*, 1107.
14. Camps, F.; Coll, J.; Riba, M. *J. Chem. Soc. Chem. Comm.*, **1979**, 1080.; Camps, F.; Coll, J.; Guerrero, A.; Guitart, J.; Riba, M. *Chem. Lett.*, **1982**, 715.
15. Akamanchi, K. G.; Patel, H. C.; Meenakshi, R. *Synth. Commun.*, **1992**, *22*, 1655.
16. Thompson, M. J.; Cohen, C. F.; Lancaster, S. M.; *Steroids*, **1965**, *5*, 745.